

# AMELIORATE EFFECT OF CALIBRIN®-Z ENTEROSORBENT ON SERUM REPRODUCTIVE HORMONE, IMMUNOGLOBULIN, ANTIBODY TITER IN YOUNG PIGS FED PURIFIED ZEARELENONE

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## ABSTRACT

Zearalenone (ZEA) is a mycotoxin that mimics estrogen; causing infertility, abortion, and other reproductive problems, especially in swine. Besides its estrogenic effects, the toxin can also negatively impact the immune response. Calibrin-Z (CAZ), a highly-refined montmorillonite sorbent mineral, has high affinity and capacity to sequester a wide range of mycotoxins found in feed grains. The objective of this study was to evaluate different dosages of CAZ on the effects of ZEA on serum hormone and immune response in piglets. A total of 36 pigs with initial body weight of  $8.9 \pm 0.2$  kg were allotted to 6 treatments (TRT). The 6 TRT were: 1) Control (Con); 2) Con + 0.1% CAZ; 3) Con + 1 ppm ZEA; 4) Con + 1 ppm ZEA + 0.1% CAZ; 5) Con + 1 ppm ZEA + 0.2% CAZ; 6) Con + 1 ppm ZEA + 0.4% CAZ. Data were analyzed using the GLM procedure of SAS with individual pig as the basis for analysis. Each pig received classical swine fever (CSF) vaccine 1-d before the study, serum samples were collected weekly for CSF antibody titer determination. On d 21, blood and spleen samples were collected for hormone, immunoglobulin, interleukin (IL) cytokine analyses, and lymphocyte proliferation rate (LPR). Gilts fed TRT 3 had lower ( $P < 0.05$ ) progesterone (75% of 1 and 2), and testosterone (74% of 1 and 2) levels than TRT 1 and 2. Adding CAZ improved serum hormone levels and the elevated hormone was CAZ dosage dependent. Male pigs fed TRT 3 showed similar results as gilts except serum estradiol was not different. Serum IgA and IgM were not different; however, IgG level was reduced ( $P < 0.01$ ) in TRT 3 (77% of 1 and 2) compared with TRT 1 and 2. It increased linearly ( $P < 0.05$ ) in TRT 4, 5, and 6 but remained lower ( $P < 0.05$ ) than TRT 1 and 2 (84% of 1 and 2). The LPR from blood and spleen cells followed a trend similar to IgG. Serum IL-2 followed results similar to those of the hormones; pigs fed TRT 3 had the lowest IL-2 but an addition of 0.4% clay restored the levels equal to TRT 1 and 2. There was no difference on d-7 or d-14 on CFS titers. On d-21, pigs fed TRT 2 had the highest titer against CSF and greater ( $P < 0.01$ ) than pigs fed TRT 3, 4, 5, and 6; but not different than TRT 1. Feeding 1 ppm of purified ZEA caused adverse effects on immunity in pigs; which were ameliorated by CAZ. Feeding CAZ without ZEA had the greatest antibody titer production, implying that the Calibrin-Z may add value to feed with or without a mycotoxin challenge.

## OBJECTIVE

The objective of this study was to evaluate the ameliorative effect of Calibrin-Z on the effects of zearalenone (ZEA) serum reproductive hormone and immune response in young pigs.

## MATERIALS & METHODS

36 weaning pigs (1/2 male, 1/2 female) Body Weight  $8.9 \pm 0.2$ kg

### Treatments

- Control
- Control + 0.1% Calibrin-Z (CZ1)
- Control + 1 ppm ZEA
- Control + 1 ppm ZEA + 0.1% Calibrin-Z (CZ1)
- Control + 1 ppm ZEA + 0.2% Calibrin-Z (CZ2)
- Control + 1 ppm ZEA + 0.4% Calibrin-Z (CZ4)

The feeding period was 21 d. Each pig received classical swine fever (CSF) vaccine 1 d before the study, serum samples were collected weekly for CSF antibody titer determination. On d 21, blood and spleen samples were collected for hormone, immunoglobulin, interleukin (IL) cytokine analyses, and lymphocyte proliferation rate (LPR). Calibrin-Z is calcium montmorillonite clay that has been further processed, resulting in increased binding of ZEA and other mycotoxins.

## RESULTS

Adding Calibrin-Z (CAZ) improved serum hormone levels and the elevated hormone was CAZ dosage dependent. Male pigs fed TRT 3 showed similar results as gilts except serum estradiol was not different. Serum IgA and IgM were not different, IgG level was reduced ( $P < 0.01$ ) in TRT 3 (77% of 1 and 2) compared with TRT 1 and 2. It increased linearly ( $P < 0.05$ ) in TRT 4, 5, and 6 but remained lower ( $P < 0.05$ ) than TRT 1 and 2 (84% of 1 and 2). The LPR from blood and spleen cells followed a trend similar to IgG. Serum IL-2 followed results similar to those of the hormones; pigs fed TRT 3 had the lowest IL-2 but an addition of 0.4% CAZ restored the levels equal to TRT 1 and 2. There was no difference on d-7 or d-14 on CFS titers. On d-21, pigs fed TRT 2 had the highest titer against CSF and greater ( $P < 0.01$ ) than pigs fed TRT 3, 4, 5, and 6; but not different than TRT 1.

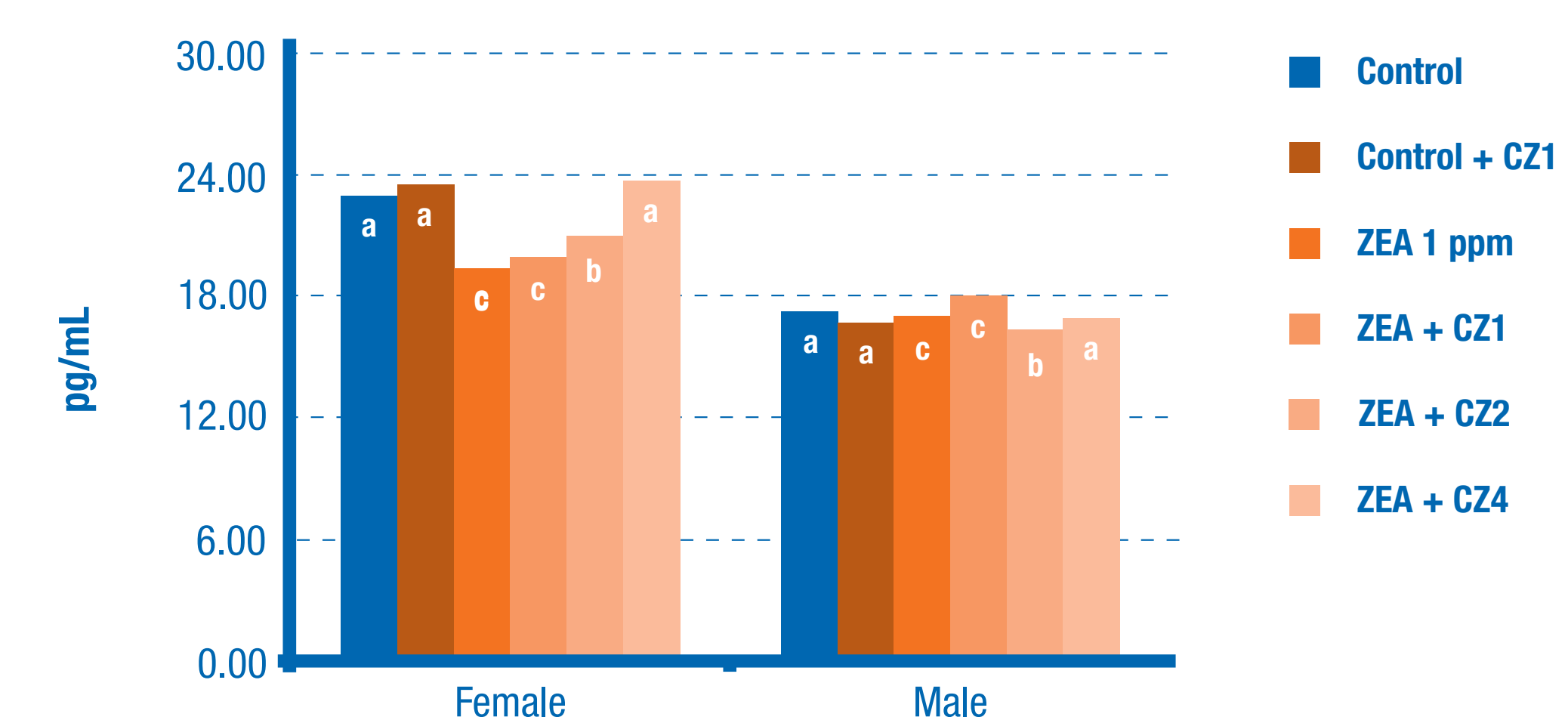
**TABLE 1.** Antibody Titer to Classic Swine Fever of Pigs Fed Zearalenone Contaminated Diets With or Without Calibrin-Z

Day From Vaccination	Control	Control + CZ1	ZEA 1 ppm	ZEA + CZ1	ZEA + CZ2	ZEA + CZ4	P values
-1	4.50	4.83	4.67	4.33	4.67	4.50	0.980
7	4.83	5.17	4.33	4.50	4.83	4.83	0.787
14	5.50	5.67	5.33	5.33	5.50	5.50	0.127
21	6.83 <sup>a</sup>	7.33 <sup>a</sup>	6.00 <sup>b</sup>	6.00 <sup>b</sup>	6.17 <sup>b</sup>	6.00 <sup>b</sup>	0.009

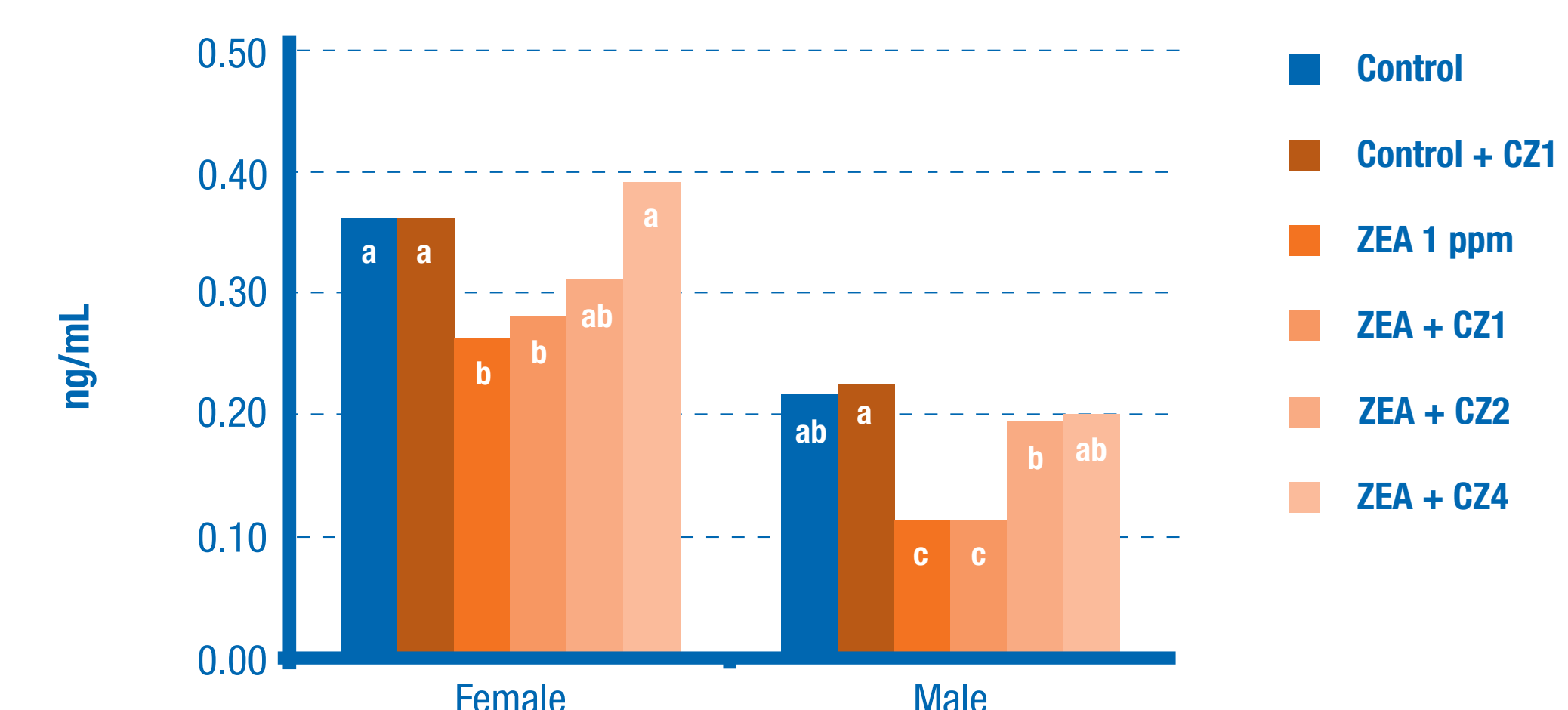
**TABLE 2.** Serum Markers of pigs Fed Zearalenone Contaminated Diets With or Without Calibrin-Z

Parameter	Control	Control + CZ1	ZEA 1 ppm	ZEA + CZ1	ZEA + CZ2	ZEA + CZ4
Aspartate aminotransferase (U/L)	68 <sup>a</sup>	66 <sup>a</sup>	86 <sup>a</sup>	80 <sup>a</sup>	76 <sup>ab</sup>	71 <sup>ab</sup>
Alanine aminotransferase (U/L)	58 <sup>a</sup>	56 <sup>a</sup>	78 <sup>a</sup>	73 <sup>a</sup>	68 <sup>ab</sup>	61 <sup>b</sup>
Alkaline phosphatase (U/L)	201 <sup>b</sup>	197 <sup>b</sup>	246 <sup>a</sup>	240 <sup>a</sup>	231 <sup>a</sup>	220 <sup>ab</sup>
γ-Glutamyltransferase (U/L)	23 <sup>a</sup>	27 <sup>a</sup>	41 <sup>a</sup>	37 <sup>a</sup>	36 <sup>a</sup>	31 <sup>ab</sup>
Urea (mmol/L)	2.7 <sup>a</sup>	2.9 <sup>b</sup>	4.1 <sup>a</sup>	3.7 <sup>a</sup>	3.4 <sup>ab</sup>	3.0 <sup>b</sup>
Creatinine (μmol/L)	77 <sup>a</sup>	75 <sup>a</sup>	88 <sup>a</sup>	91 <sup>a</sup>	79 <sup>ab</sup>	75 <sup>b</sup>

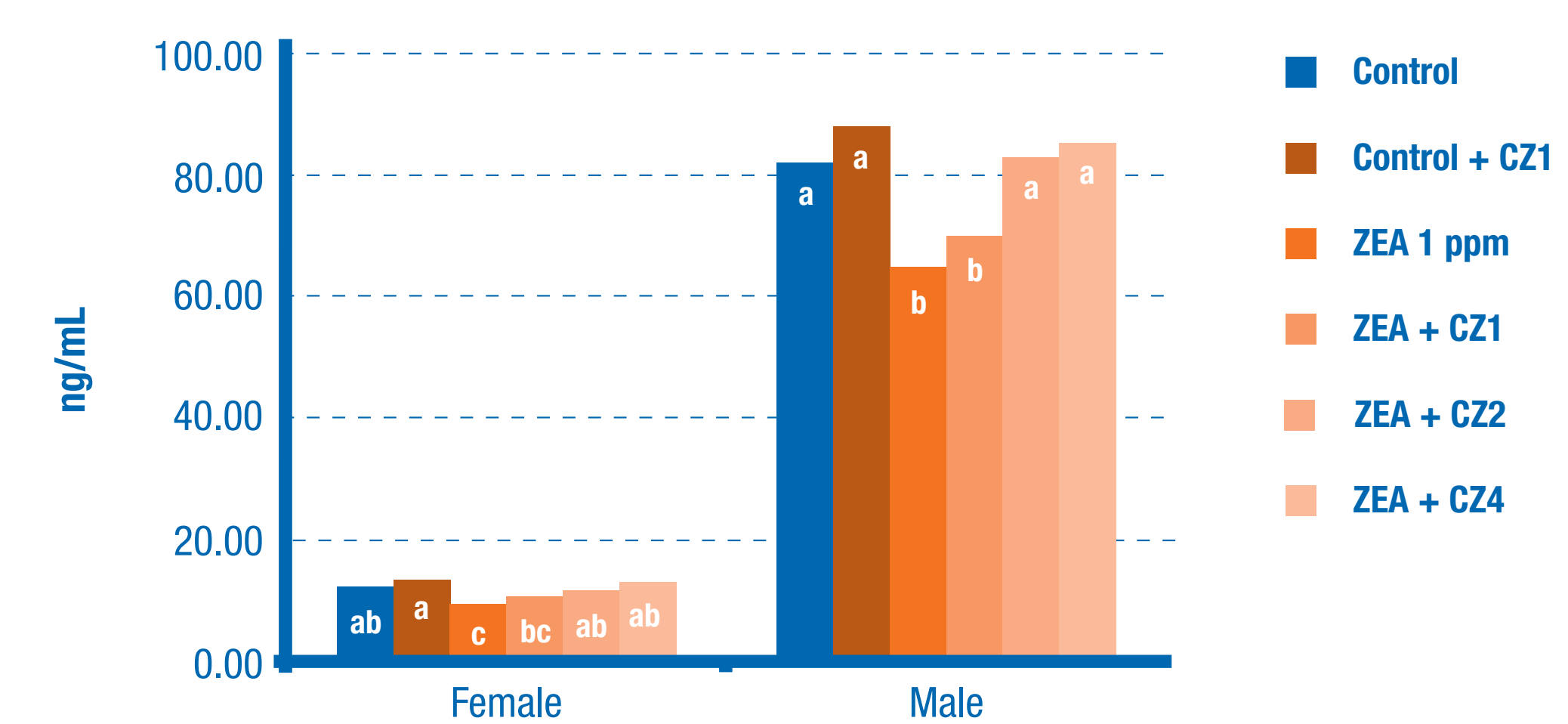
**FIGURE 1.** Serum estradiol of pigs fed zearalenone contaminated diets with or without Calibrin-Z



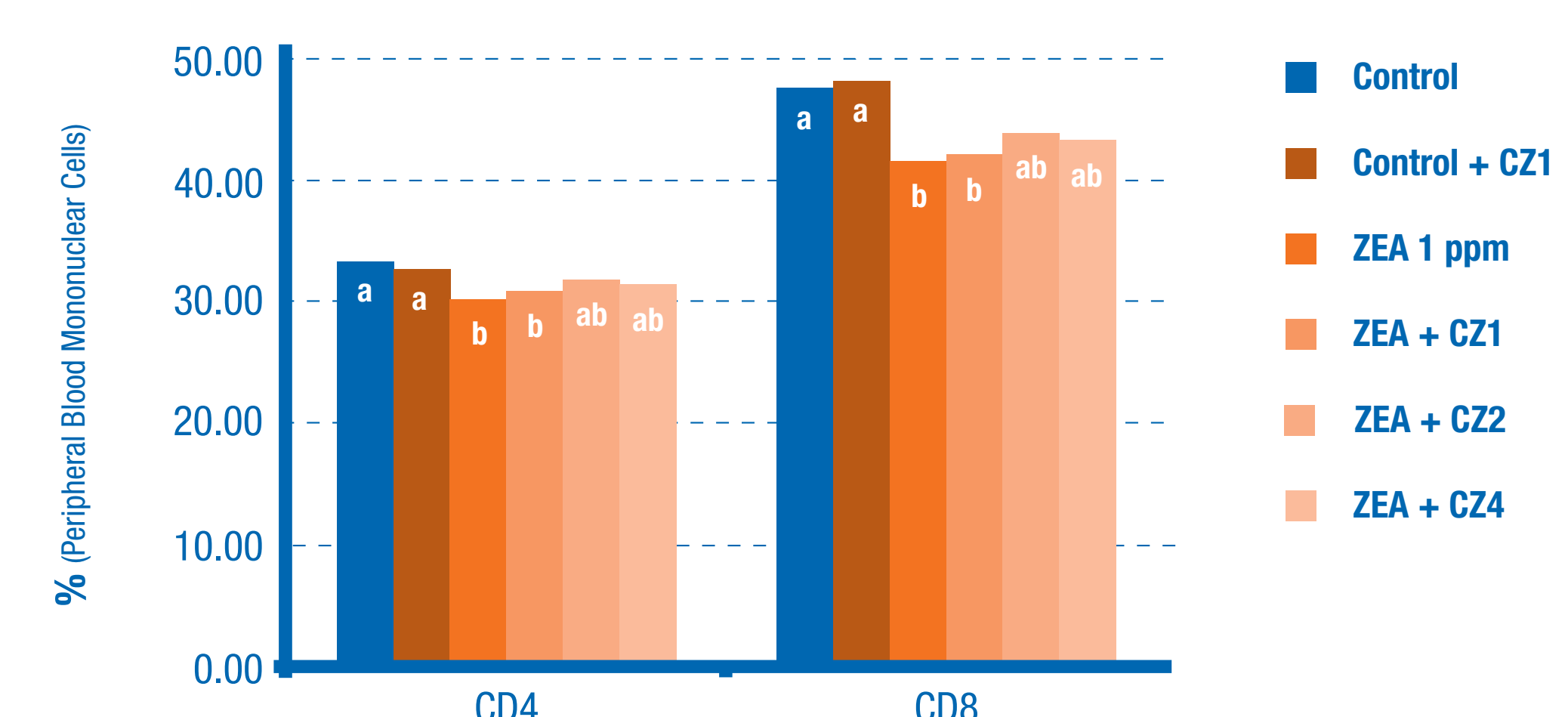
**FIGURE 2.** Serum progesterone of pigs fed zearalenone contaminated diets with or without Calibrin-Z



**FIGURE 3.** Serum testosterone of pigs fed zearalenone contaminated diets with or without Calibrin-Z



**FIGURE 4.** CD4 and CD8 T-cell counts of pigs fed zearalenone contaminated diets with or without Calibrin-Z



## CONCLUSIONS

- Feeding 1 ppm of purified ZEA caused adverse effects on immunity in pigs; which were ameliorated by Calibrin-Z.
- Feeding Calibrin-Z without ZEA had the greatest antibody titer production, implying that the Calibrin-Z adds value to feed with or without a mycotoxin challenge.